REMARKS

Present Invention and Pending Claims

Claims 1, 4, and 6 are pending and directed to a plasmid defective for conjugative transfer function comprising a DNA fragment containing a gene coding glucose dehydrogenase (claim 1), a bacterial transformant comprising the plasmid (claim 4), and a method of producing glucose dehydrogenase (claim 6).

Amendments to the Claims

The claims have been amended to point out more particularly and claim more distinctly the present invention. Specifically, claim 1 has been amended to incorporate the features of claims 2 and 3. Accordingly, claims 2 and 3 have been canceled to prevent redundancy. Claim 4 has been amended to recite that the enzyme is glucose dehydrogenase and that the plasmid is introduced into a bacterial strain of the genus *Pseudomonas*. Claim 4 has amended to incorporate the features of claim 5. Accordingly, claim 5 has been canceled to prevent redundancy. The amendments to claim 4 are supported by the specification at, for example, page 4, lines 24-26, and page 8, lines 20-29. Claim 6 has been amended to recite that the enzyme is glucose dehydrogenase. The amendment to claim 6 is supported by the specification at, for example, page 8, lines 20-29. Accordingly, no new matter has been added by way of these amendments.

The Office Action

The Office has rejected claims 1-6 under 35 USC § 103(a) as being allegedly obvious in view of Takeshima et al. (JP 11-243949) in view of Cameron et al. (U.S. Patent 5,670,343). Reconsideration of the rejection is hereby requested.

Discussion of the Rejection

The Examiner contends that one of ordinary skill in the art would have been motivated to make pGLD3 a non-mobilizable vector by mutating or deleting the mob locus. The Examiner contends that the Cameron reference teaches that plasmids that are non-conjugative and non-mobilizable are preferred for use in *E. coli* and *Psuedomonas* for improved biosafety (e.g., to prevent transmission to other organisms). Thus, the Examiner contends that the Cameron reference provides the motivation and the means to combine the teachings of the Cameron reference with the Takeshima reference. Applicants traverse this rejection for the following reasons.

The inventors unexpectedly discovered that the expressed activity of glucose dehydrogenase produced by a plasmid lacking conjugative transfer function, as assayed by the amount of enzyme produced by the plasmid, is comparable to, or higher than, the expressed activity of glucose dehydrogenase produced by a plasmid retaining conjugative transfer function. For example, the data in Table 2 of the specification indicates that 27 U/mL of expressed activity was recorded for *P. putida* TN1126/pGLD6 (pGLD6 lacks conjugative transfer function). The data in Table 1 of the Takeshima reference indicates that 26 U/mL of expressed activity was recorded for *P. putida* TN1126/pGLD3, wherein pGLD3 differs from pGLD6 solely by retaining conjugative transfer function (mob genes).

One of ordinary skill in the art would not have expected that the removal of the mob locus (as disclosed in the Cameron reference) would have resulted in an expressed activity of glucose dehydrogenase produced by the plasmid lacking conjugative transfer function comparable to, or higher than, that of a plasmid retaining conjugative transfer function. Since the genes relating to replication, such as rep A, rep B, and rep C, as well as ori T, which is the starting point of the rep genes, are located near the mob locus, one of ordinary skill in the art would have expected that the removal or mutation of the mob locus could have a negative effect on the genes surrounding the mob locus. For example, when mob genes are cleaved from vectors, portions of the DNA backbone near the mob genes often also are deleted as a result of experimental conditions, as well as the restriction enzymes. With the plasmid of the Takeshima reference, the deletion of a portion of the DNA backbone near the mob gene could mean that the rep genes would be impaired, thereby causing a lower replication efficiency of the vector and lower expressed activity of the encoded enzyme.

Accordingly, one of ordinary skill in the art would not have expected that the removal of the conjugative transfer function in the plasmid of the Takeshima reference by deletion of a region carrying the mob locus in accordance with the Cameron reference would result in comparable, if not higher, expressed activity of the glucose dehydrogenase as compared to a plasmid retaining conjugative function (i.e., the mob locus). There is nothing in either the Takeshima reference or the Cameron reference that suggests that removal or mutation of the mob locus would result in an expressed activity of an enzyme encoded by a plasmid that lacks conjugative function that is comparable to, or higher than, that of a plasmid that retains conjugative function.

Under the circumstances, and especially in view of the unexpected results attendant the present invention, the present invention as defined by the pending claims must be considered unobvious over the disclosures of the Takeshima and Cameron references. As such, the Section 103(a) rejection should be withdrawn.

Conclusion

The application is considered in good and proper form for allowance, and the Examiner is respectfully requested to pass this application to issue. If, in the opinion of the Examiner, a telephone conference would expedite the prosecution of the subject application, the Examiner is invited to call the undersigned attorney.

Respectfully submitted,

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